

## SEQUENCE LISTING

<110> Happe, Scott B.

Leininger, Katie J.

Dubois, Dwight B.

<120> Humanized Renilla Reniformis Green Fluorescent Protein As A Scaffold

<130> 25436/2282

<140> Not yet assigned

<141> 2003-07-10

<150> US 60/394,737

<151> 2002-07-10

<160> 12

<170> PatentIn version 3.1

<210> 1

<211> 720

<212> DNA

<213> Artificial

<220>

<221> misc\_feature

<222> (1) .. (720)

<223> Humanized version of Renilla reniformis Green Fluorescent Protein

coding sequence

<400> 1  
atggtgagca agcagatcct gaagaacacc tgcctgcagg aggtgatgag ctacaagggtg 60  
aacctggagg gcatcgtgaa caaccacgtg ttcaccatgg agggctgcgg caagggcaac 120  
atcctgttcg gcaaccagct ggtgcagatc cgcgtgacca agggcgcccc cctgcccttc 180  
gccttcgaca tcgtgagccc cgccttccag tacggcaacc gcaccttcac caagtacccc 240  
aacgacatca gcgactactt catccagagc ttccccgccg gcttcatgta cgagcgcacc 300  
ctgcgctacg aggacggcgg cctggtggag atccgcagcg acatcaacct gatcgaggac 360  
aagttcgtgt accgcgtgga gtacaagggc agcaacttcc ccgacgacgg ccccgatgatg 420  
cagaagacca tcctgggcat cgagcccagc ttcgaggcca tgtacatgaa caacggcgtg 480  
ctggtgggcg aggtgatcct ggtgtacaag ctgaacagcg gcaagtacta cagctgccac 540  
atgaagaccc tgatgaagag caagggcgtg gtgaaggagt tcccctccta ccacttcac 600  
cagcaccgcc tggagaagac ctacgtggag gacggcggct tcgtggagca gcacgagacc 660  
gccatcgccc agatgaccag catcggcaag cccctgggca gcctgcacga gtgggtgtaa 720

<210> 2

<211> 738

<212> DNA

<213> Artificial

<220>

<221> misc\_feature

<222> (1)..(738)

<223> Humanized version of Renilla reniformis Green Fluorescent Protein  
coding sequence with 18 bp insert.

<400> 2  
atggtgagca agcagatcct gaagaacacc ggcctgcagg agatcatgag cttcaagggtg 60  
aacctggagg gcgtggtgaa caaccacgtg ttcaccatgg agggctgcgg caagggcaac 120  
atcctgttcg gcaaccagct ggtgcagatc cgcgtgacca agggcgcccc cctgcccttc 180

gccttcgaca tcctgagccc cgccttccag tacggcaacc gcaccttcac caagtacccc 240  
gaggacatca gcgacttctt catccagagc ttccccgccg gcttcgtgta cgagcgcacc 300  
ctgcgctacg aggacggcgg cctggtggag atccgcagcg acatcaacct gatcgaggag 360  
atgttcgtgt accgcgtgga gtacaagggc cgcaacttcc ccaacgacgg ccccgatgat 420  
aagaagacca tcaccggcct gcagcccagc ttcgaggtgg tgtacatgaa cgacggcgtg 480  
ctggtggggc aggtgatcct ggtgtaccgc ctgaacagca gatctgaatt cgacgtcggc 540  
aagttctaca gctgccacat gcgcaccctg atgaagagca agggcgtggt gaaggacttc 600  
cccagtagtacc acttcatcca gcaccgcctg gagaagacct acgtggagga cggcggcttc 660  
gtggagcagc acgagaccgc catcgcccag ctgaccagcc tgggcaagcc cctgggcagc 720  
ctgcacgagt ggggtgtaa 738

<210> 3

<211> 239

<212> PRT

<213> Renilla reniformis

<400> 3

Met Val Ser Lys Gln Ile Leu Lys Asn Thr Gly Leu Gln Glu Ile Met  
1 5 10 15

Ser Phe Lys Val Asn Leu Glu Gly Val Val Asn Asn His Val Phe Thr  
20 25 30

Met Glu Gly Cys Gly Lys Gly Asn Ile Leu Phe Gly Asn Gln Leu Val  
35 40 45

Gln Ile Arg Val Thr Lys Gly Ala Pro Leu Pro Phe Ala Phe Asp Ile  
50 55 60

Leu Ser Pro Ala Phe Gln Tyr Gly Asn Arg Thr Phe Thr Lys Tyr Pro  
65 70 75 80

Glu Asp Ile Ser Asp Phe Phe Ile Gln Ser Phe Pro Ala Gly Phe Val  
85 90 95

Tyr Glu Arg Thr Leu Arg Tyr Glu Asp Gly Gly Leu Val Glu Ile Arg  
100 105 110

Ser Asp Ile Asn Leu Ile Glu Glu Met Phe Val Tyr Arg Val Glu Tyr  
115 120 125

Lys Gly Arg Asn Phe Pro Asn Asp Gly Pro Val Met Lys Lys Thr Ile  
130 135 140

Thr Gly Leu Gln Pro Ser Phe Glu Val Val Tyr Met Asn Asp Gly Val  
145 150 155 160

Leu Val Gly Gln Val Ile Leu Val Tyr Arg Leu Asn Ser Gly Lys Phe  
165 170 175

Tyr Ser Cys His Met Arg Thr Leu Met Lys Ser Lys Gly Val Val Lys  
180 185 190

Asp Phe Pro Glu Tyr His Phe Ile Gln His Arg Leu Glu Lys Thr Tyr  
195 200 205

Val Glu Asp Gly Gly Phe Val Glu Gln His Glu Thr Ala Ile Ala Gln  
210 215 220

Leu Thr Ser Leu Gly Lys Pro Leu Gly Ser Leu His Glu Trp Val  
225 230 235

<210> 4

<211> 245

<212> PRT

<213> Artificial

<220>

<221> MISC\_FEATURE

<222> (1)..(245)

<223> Renilla reniformis GFP with 6 amino acid insert encoded by the 18  
base pair insert in SEQ ID NO: 2.

<400> 4

Met Val Ser Lys Gln Ile Leu Lys Asn Thr Gly Leu Gln Glu Ile Met  
1 5 10 15

Ser Phe Lys Val Asn Leu Glu Gly Val Val Asn Asn His Val Phe Thr  
20 25 30

Met Glu Gly Cys Gly Lys Gly Asn Ile Leu Phe Gly Asn Gln Leu Val  
35 40 45

Gln Ile Arg Val Thr Lys Gly Ala Pro Leu Pro Phe Ala Phe Asp Ile  
50 55 60

Leu Ser Pro Ala Phe Gln Tyr Gly Asn Arg Thr Phe Thr Lys Tyr Pro  
65 70 75 80

Glu Asp Ile Ser Asp Phe Phe Ile Gln Ser Phe Pro Ala Gly Phe Val  
85 90 95

Tyr Glu Arg Thr Leu Arg Tyr Glu Asp Gly Gly Leu Val Glu Ile Arg  
100 105 110

Ser Asp Ile Asn Leu Ile Glu Glu Met Phe Val Tyr Arg Val Glu Tyr  
115 120 125

Lys Gly Arg Asn Phe Pro Asn Asp Gly Pro Val Met Lys Lys Thr Ile  
130 135 140

Thr Gly Leu Gln Pro Ser Phe Glu Val Val Tyr Met Asn Asp Gly Val  
145 150 155 160

Leu Val Gly Gln Val Ile Leu Val Tyr Arg Leu Asn Ser Arg Ser Glu  
165 170 175

Phe Asp Val Gly Lys Phe Tyr Ser Cys His Met Arg Thr Leu Met Lys  
180 185 190

Ser Lys Gly Val Val Lys Asp Phe Pro Glu Tyr His Phe Ile Gln His  
195 200 205

Arg Leu Glu Lys Thr Tyr Val Glu Asp Gly Gly Phe Val Glu Gln His

210

215

220

Glu Thr Ala Ile Ala Gln Leu Thr Ser Leu Gly Lys Pro Leu Gly Ser  
 225 230 235 240

Leu His Glu Trp Val  
 245

&lt;210&gt; 5

&lt;211&gt; 720

&lt;212&gt; DNA

&lt;213&gt; Renilla reniformis

&lt;400&gt; 5

atggtgagta aacaaatatt gaagaacact ggattgcagg agatcatgtc gtttaaagtg 60  
 aatctggaag gtgtagtaaa caatcatgtg ttcacaatgg aaggttgtgg aaaaggaaat 120  
 attttattcg gaaaccaact gggtcagatt cgtgtcacaa aaggggctcc gcttccattt 180  
 gcatttgata ttctctcacc agctttccaa tacggcaacc gtacattcac gaaataccgc 240  
 gaggatatat cagacttttt tatacaatca tttccagcgg gatttgtata cgaaagaacg 300  
 ttgcgttacg aagatggtgg actggttgaa atccgttcag atataaattt aatcgaggag 360  
 atgtttgtct acagagtgga atataaaggt agtaacttcc cgaatgatgg tccagtgatg 420  
 aagaagacaa tcacaggatt acaaccttcg ttcgaagttg tgtatatgaa cgatggcgtc 480  
 ttggttgggc aagtcattct tgtttataga ttaaactctg gcaaatttta ttcgtgtcac 540  
 atgagaacac tgatgaaatc aaaggggtgta gtgaaggatt ttcccgaata ccatttcatt 600  
 caacatcgtt tagagaagac gtatgtggaa gacggaggtt ttgttgagca acacgagacg 660  
 gccattgctc aactgacatc gctggggaaa ccacttgat ccttacacga atgggtttta 720

&lt;210&gt; 6

&lt;211&gt; 44

&lt;212&gt; DNA

&lt;213&gt; Artificial

<220>

<221> misc\_feature

<222> (1)..(44)

<223> Forward PCR Primer to amplify R. reniformis GFP, including artificial EcoRI site and Kozak consensus.

<400> 6

aattattaga attcaccatg gtgagtaaac aaatattgaa gaac

44

<210> 7

<211> 38

<212> DNA

<213> Artificial

<220>

<221> misc\_feature

<222> (1)..(38)

<223> Reverse PCR primer for Renilla reniformis GFP, including artificial XhoI site.

<400> 7

ataatattct cgagttaaac ccattcgtgt aaggatcc

38

<210> 8

<211> 6

<212> PRT

<213> Renilla reniformis

<400> 8

Phe Gln Tyr Gly Asn Arg

1

5

<210> 9

<211> 39

<212> DNA

<213> Artificial

<220>

<221> misc\_feature

<222> (1)..(39)

<223> Synthetic PCR primer used in construction of hrGFP-173

<400> 9

attattgcgg ccgcatccac catggtgagc aagcagatc

39

<210> 10

<211> 39

<212> DNA

<213> Artificial

<220>

<221> misc\_feature

<222> (1)..(39)

<223> Synthetic PCR primer used in the construction of hrGFP-173.

<400> 10

attattgaat tcgacgtcgg caagttctac agctgccac

39

<210> 11

<211> 38

<212> DNA

<213> Artificial



<220>

<221> misc\_feature

<222> (1)..(38)

<223> Synthetic PCR primer used in construction of hrGFP-173.

<400> 11

attattgaat tcagatctgc tggtcaggcg gtacacca

38

<210> 12

<211> 37

<212> DNA

<213> Artificial

<220>

<221> misc\_feature

<222> (1)..(37)

<223> Synthetic PCR primer used in construction of hrGFP-173.

<400> 12

attattattc tcgagctatt acaccactc gtgcagg

37